THE EFFECT OF TOTAL BODY IRRADIATION ON IMMUNOLOGIC
TOLERANCE OF BONE MARROW AND HOMOGRAFTS OF
OTHER LIVING TISSUE

A GOVERNMENT RESEARCH REPORT

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Nuclear Weapons Effects Research No. 03.011

Sub-task Title: The Effect of Total Body Irradiation on Immunoiogic
Tolerance of Bone Marrow and Homografts of
Other Living Tissue

Service Project No.: DA-49-146-XZ-032
Modification No. 3

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Classification: Unclassified

Report Feriod: Feb. 1, 1963 - Jan. 31, 1964
Final Report

Investigations in the effect of total body irradiation have been carried out in this department for more than 10 years and this final report is based on data collected during this time. The project was initiated when comparison of response of patients receiving therapeutic total body irradiation with that of patients receiving chemotherapy indicated that, in the dose range used, total body raidation was well tolerated and could be used as as adjunct to treatment of disseminated cancer (1, 2). Intensive studies on response of erythropoiesis were undertaken (3) and findings gave new directions for further study so that, in the final years, the protocol included a large series of clinical, hematologic, biochemical and physiological tests (4) as well as studies in protection. The investigation includes observations on 112 patients receiving therapeutic total body irradiation; pilot and parallel studies were carried out on experimental animals.

The fundamental problem has been to define effect of irradiation and quantitate effect with amount of radiation exposure. Since quantitation is possible only to the degree that measurement of both effect and amount of radiation is possible, it was necessary to consider all the variables that would influence measurement.

Comprehension of the problem begins with the recognition that a simple numerical expression cannot define dose of irradiation. The physical characteristics of irradiation require that dose be described not only by the number of units (roentgens, rads, etc.) but also by distribution and intensity in a given volume of tissue, and time or duration of exposure. For each irradiation procedure planned, dosimetry studies were carried out so that uniform distribution of irradiation could be achieved. Several methods, and types of dos'meters, were used for comparison, and for accurate measurement of dose to specific anatomical sites. These methods assured even distribution of radiation and produced comparable data on radiation exposure throughout the entire study.

Similar dosimetry studies were carried out for animal experiments. Protocols required animals of similar age, weight and physical structure so that volume irradiated and response could be compared. All animals received immunization vaccine, were quarantined for two weeks, and all were well nourished and in good health prior to experimental work.

Although variables associated with patients requiring therapeutic total body irradiation are present, certain aspects were comparable. All patients were adults suffering from disseminated cancer, all received evenly distributed exposures of irradiation and, except when reaction was severe, none received supportive treatment that would mask effect of irradiation. Since prediction of effect is the basis of radiotherapy, experience in this field is helpful in evaluating response to treatment and in estimating response to heavy exposures by extrapolation of data obtained with therapeutic exposures.

Therapeutic Total Body Irradiation

Examinations and laboratory tests were done on all patients prior to therapeutic total body irradiation to obtain a base line for comparison with tests made during and following treatment. Records of all clinical findings, results of all tests, factors, technique and dose of irradiation were maintained. Follow-up continued as long as the patient survived and, when possible, autopsy was obtained to determine whether or not there was evidence of radiation change. These records formed the basis for analyses of changes by various parameters as presented in annual progress reports.

The patients were divided into 3 groups according to duration (elapsed time) of radiation exposure. These are defined as: I - Single Exposure, II - Protracted Irradiation (given within a period of 2 months), and III - Repeated courses (period ranging from 3 months to 3½ years). Because tolerance to radiation increases with protraction of time, there is a tendency to emphasize protraction while overlooking the amount that can be given safely in a single exposure. When nearing the terminal stage of disease, few patients are likely to survive a protracted course of therapy and dose, which is necessarily low for each treatment of a protracted course, is not likely to prove beneficial in the brief time available for therapeutic effect. Thus, the majority of patients (71/112 - 63%) in this series received single exposures. It seemed possible that response in this group might be compared with data obtained from study of accident victims and exposed Japanese populations who received single exposures.

Group II is composed of 34 (30%) patients receiving protracted irradiation. These were selected because of generalized disease that might be expected to respond to small repeated exposures. It was expected that study of this group would provide information concerning response that might be useful among military personnel when occupying a radioactive area.

The third group is composed of 7 patients who had received either single or protracted exposures and who, after months or years of remission of disease, developed recurrent symptoms requiring further treatment by total body irradiation. These 7 patients received a total of 16 separate courses of treatment or 9 studies in addition to the 112 patients. The number of patients, amount and range of irradiation exposure, and elapsed time, is summarized in Table 1.

Table 1

Category	Patients	Range of Exposure and Time
I.	71 (63%)	25r - 250r/1 day
II.	34 (31%)	25r/4 days - 400r/32 days
III.	7 (6%)	50r-250r/5 mos. - 2 courses
		50r-200r/3½ yrs. - 4 courses

Table 2 summarizes the number of patients receiving total body irradiation in a single exposure. Amount of exposure ranged from 25r to 250r, depending upon the therapeutic indications for treatment.

Table 2

No. of Patients	Total Body Irradiation (single exposure)
6	25r
20	50r
5	75r
8	100r
1	125r
7	150r
22	200r
1	215r
1	250r

Table 3 summarizes the number of patients receiving therapeutic total body irradiation, protracted over periods ranging from 2 to 33 days.

Table 3

No. of Patients	Amount of Irradiation	Elapsed Time
1	25r	4 days
2	50r	2 - 5 days
2	100r	7 - 28 days
9	150r	3 - 18 days
1	185r	7 days
4	200r	6 - 19 days
1	215r	21 days
6	250r	7 - 17 days
4	300r	6 - 33 days
2	350r	22 days
1	400r	23 days
1	545r	18 days

Table 4 outlines the amount of radiation with duration of exposure and time lapse between each course for the 7 patients receiving repeated courses of therapeutic total body irradiation.

Table 4

Patients	Time Lapse	Total Time	Amount of Radiation
M.M.	0		125r/62 days
	8 mos.		50r/11 days
	17 mos.	25 mos.	282r/113 days
F.M.	0		200r/1 day
	5 mos.	5 mos.	200r/1 day
I.S.	0		90r/35 days
	3 mos.		30r/16 days
	1 mo.	4 mos.	50r/63 days
E.R.	0		100r/35 days
	4 mos.		50r/14 days
	9 mos.		150r/1 day
	29 mos.	42 mos.	200r/9 days
M.B.	0		150r/1 day
	10 mos.		100r/1 day
	7 mos.	17 mos.	50r/1 day
J.B.	0		250r/58 days
	5 mos.	5 mos.	50r/1 day
J.T.	0		100r/7 days
	25 mos.	25 mos.	100r/6 days

Systemic reaction following therapeutic total body irradiation is difficult to evaluate because of the influence of disease, medication, and psychogenic factors. Malaise following irradiation was recognized and reported as early as 1897 but the train of symptoms, in 1912 termed "Rontgenkater" (roentgen-hangover), for a given amount of radiation varies widely. Ellinger summarized a series of biochemical changes (5) which he believed accounted for "radiation sickness." Since symptoms may appear following exposures too light to produce detectable biochemical changes, and may not develop following marked changes, other factors must be considered.

For evaluation of the present series, symptoms were graded according to severity: 0 = no symptoms, 1 = anorexia only, 2 = nausea only, 3 = nausea and vomiting one day, and 4 = nausea and vomiting more than one day. Two patients are excluded from this study; one, terminal at the time of treatment, died within 24 hours, and one, an out-patient with concurrent tuberculosis, was lost to follow-up. Table 5 summarizes the severity of symptoms according to single, protracted and repeated courses of exposure among 110 patients.

Table 5

Radiation Exposure	No. of Patients	Symptoms (grades)				
		0	1	2	3	4
I- Single exposure	69	54	4	3*	8*	0
II- Protracted exposures	34	29	2	2	1	0
III- Repeated courses	7	6	0	0	0	1
Total	110	89	6	5	9	1

*One patient with Grade 2 and one patient with Grade 3 symptoms had nausea and vomiting prior to irradiation.

A total of 21 patients (19%) developed reactions varying from mild to severe but this cannot be accepted as an indication of incidence that might occur in an exposed population. In this group any or all symptoms could be due to disease (2 patients had symptoms prior to treatment), but there is no clue to explain why 89 patients (81%) developed no symptoms at all.

In evaluating radiation sickness according to amount of irradiation, patients receiving single exposures are studied because response is easily recognized and is not complicated by cumulative effect of repeated exposures. Table 6 shows the reaction of 69 patients receiving single exposures of therapeutic total body irradiation

Table 6

Amount of Irradiation	No. of Patients	Symptoms (grades)				
		0	1	2	3	4
25r	5	5	0	0	0	0
50r	20	15	0	2	3	0
75r	5	5	0	0	0	0
100r	8	8	0	0	0	0
125r	1	1	0	0	0	0
150r	7	5	2	0	0	0
200r	21	15	2	1	3*	0
215r	1	0	0	0	1	0
250r	1	0	0	0	1*	0
Total	69	54	4	3	8	0

*One patient receiving 200r and one receiving 250r had nausea and vomiting prior to irradiation.

Study of the table shows the incidence of symptomatic response to total body irradiation is practically the same for patients receiving 50r as for those receiving 200r, but 14 patients receiving 75 to 125r failed to develop complaints. Review of the clinical histories of the 5 patients with symptoms following 50r reveals that 4 were occupants of the same ward at the same time. The subject of radiation sickness had been publicized in the popular press, was discussed by all occupants of the

ward and anticipated by the 4 scheduled for treatment. The 5th patient, terminal at the time of treatment, was receiving heavy doses of narcotics for pain from widespread involvement of disease from primary breast cancer; in this case, symptoms could have been caused by disease, medication, motion and manipulation, or a combination of these factors.

A careful study of all patients who developed symptoms following therapeutic total body irradiation indicates that, for levels up to 200r in single or protracted exposures, radiation sickness may be avoided by proper management. The attitude and experience of professional personnel, the intentions of personal acquaintances in contact with the patient, the reliability of information in the popular press, and the stability of the patient,--all these enter into the problem of motivation or suggestion. It is probable that, with correct information and proper preparation, normal healthy individuals could tolerate even higher exposures without undue incapacitation.

The goal of treatment by total body irradiation for patients with disseminated cancer is relief of symptoms. Depsite the fact that many patients were approaching the terminal stage when referred for treatment, therapeutic response was evident in many by decrease in size of nodes, relief of impending obstruction and by decreased narcotic requirements subsequent to relief of pain. In some instances, response was dramatic; a few completely bedridden patients became ambulatory and several experienced long term remissions. Among the 112 patients there were more than 30 different types of cancer; since total body irradiation was given only for generalized disease, a survival of one year or even temporary remission was gratifying. In Table 7, the diagnoses are shown in 17 related groups with the number of patients in each group.

Table 7

Diagnosis	No. of Patients
Leukemia (chronic & acute)	14
Lymphosarcoma	23
Lung cancer	12
Hodgkin's disease	6
Multiple myeloma	9
Head & Neck (larynx, tongue, etc.)	7
Testicular cancer	5
Breast cancer	3
G.I. tract (stomach, pancreas)	4
Gynecological cancer	2
Mycosis fungoides	2
Ewing's sarcoma	2
Carcinomatosis (primary unknown)	5
Miscellaneous cancer (paraganglioma, cancer of liver, kidney, prostate, & malignant melanoma)	14
Total	112

Among patients with far advanced cancer, effect of treatment cannot be measured in terms of survival but survivals, through the expected period of depression and recovery, provide information on immediate biologic and physiologic response to total body irradiation. Table 8 summarizes the survival of all patients in the study.

Table 8

Survival Time	Number of Patients		
	Died	Lost to F.U.	Living
0 - 3 mos.	47	6	0
3 - 6 mo.	21	0	0
6 - 12 mos.	10	0	0
1 - 2 yrs.	11	0	0
2 - 3 yrs.	4	0	0
3 - 4 yrs.	0	1	1
4 - 5 yrs.	0	0	0
5 - 6 yrs.	0	1	1
6 - 7 yrs.	1	2	0
7 - 8 yrs.	1	1	3
8 - 9 yrs.	0	0	0
9 - 10 yrs.	0	0	0
10 - 11 yrs.	0	0	2
	95	10	7

Of the 95 patients known to have died, 93 died of disease within 3 years following treatment. Ten patients (9%) were lost to follow-up but 4 of these were observed for periods ranging from 3 to 7 hears before they were lost. Seven patients are living with survival ranging from 3 to 10½ years.

Although patients with disseminated disease from many types of cancer received therapeutic total body irradiation (see Table 7), greatest benefit is derived by those with lymphoma, manifested chiefly in enlarged lymph nodes without marked bone marrow involvement. Over the past 10 years, the increasing activities of the Chemotherapeutic Trials Committee have reduced the number of referrals of such patients for radiotherapy to practically zero. Thus, the majority of patients are those who failed to respond to chemotherapy or those who were refused chemotherapy because of anemia and poor general condition. Among cancer patients, anemia may be caused by infiltration of cancer cells in bone marrow with replacement of normal pre-cursor cells and by shortened red cell survival (6). With widespread cancer, anemia, whether frank or subclinical, is present and death within a short time is inevitable. When patients are referred as a "last resort," the radiotherapist does not wish to withhold treatment that may offer possible benefit but he cannot be certain that the benefit will outweigh the risk. The risk is not that the patient will die but that the undesirable effects of radiation will appear more severe in the terminal cancer patient and that the time of death may be destined to coincide with the undesirable effects of radiation. Supportive treatment, even though

it may not influence the course of disease, must be included in the plan of treatment for such patients. The following case history illustrates some of the problems involved.

A.S. This 73 year old lawyer was admitted with history of hacking, productive cough, left chest pain, fever 102° and generalized lymphadenopathy of 7 weeks duration.

| Date	Day	
2/20/63		On admission, examination revealed bilateral cervical, supraclavicular, axillary & inguinal nodes 1 - 3 cm. in diameter; liver 4 cm. and spleen 3 cm. below the costal margin. Chest x-ray - enlarged hilar nodes; lymphangiogram demonstrated enlarged pelvic nodes, bilateral, with filling defects & replacement of normal architecture by tumor. Urine & sputum culture - positive; tetracycline 250 mg. q.i.d. Biopsy - lymphosarcoma, lymphocytic cell type. Bone marrow: diagnosis - diffusely involved by tumor, 275 cc. stored. Hgb. 9.2; WBC 4,500.
3/15/63		Antibiotics continued with little benefit; nodes increasing in size. Hgb. 6.8; WBC 4,100: Plts. 192,000
3/16/63	0	Total body irradiation 185r/6 days, 2 m v No nausea or vomiting
3/21/63	5	Hgb. 6.8; WBC 2,500. Transfusion 500 cc. whole blood to combat infection; 50% regression in size of nodes and masses.
4/2/63	17	Hgb. 8.8; WBC 1,100; Plts. 190,000
4/6/63	21	Sputum still positive; Rx colymycin, isolation
4/8/63	23	Re-infusion of marrow not feasible Transfusion - 1000 cc. whole blood
4/12/63	27	Hgb. 11.2; WBC 177; Plts. 28,000
4/14/63	29	Expired.

Comment. Anemia and infection were present on admission and treatment was delayed for nearly a month while an attempt was made to improve the patient's general condition. During this time, nodes and masses continued to enlarge, anemia progressed, and it was evident that there was no chance for improvement unless treatment for lymphosarcoma was offered. Marked regression of nodes indicated good therapeutic response but this was overshadowed by bone marrow depression in the presence of extensive bone marrow involvement and infection. Figure 1 shows response of white blood cell count compared with response of total circulating granulocytes.

Although 68 patients died of disease within 6 months following therapeutic total body irradiation, many of these experienced temporary remission and contributed valuable information to the study. The following case history is an example of good therapeutic effect providing opportunity to observe response through the period of bone marrow depression and recovery.

Fig. 1.

Total Body Irradiation

(A.S.) Comparison of response of white blood cell count and
granulocytes following 185r/6 days
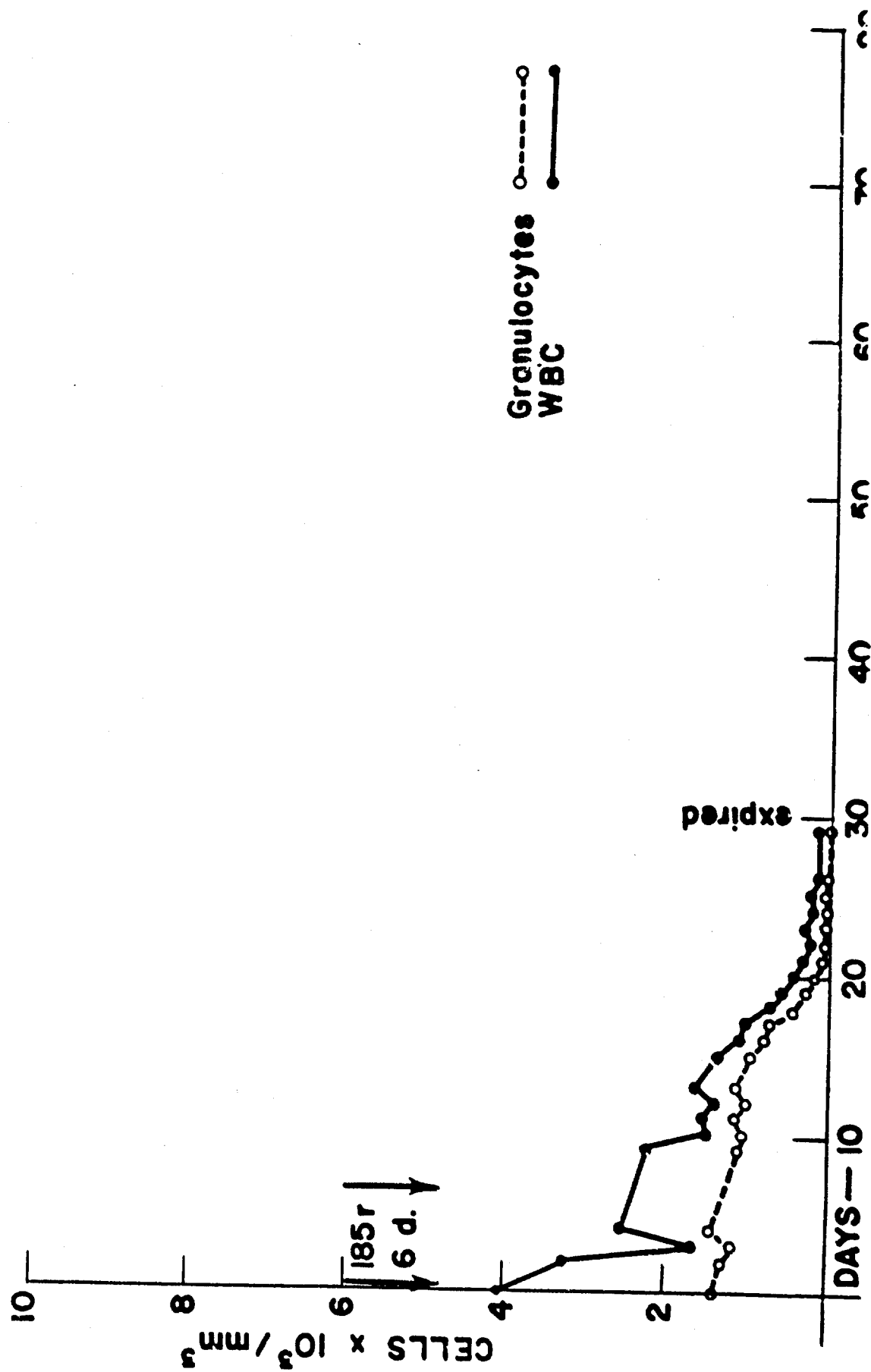
J.C. This 37 year old man was admitted complaining of epigastric
pain with hematemesis of 5 years duration; jaundice with enlarged
liver (7 cm. below costal margin) and retroperitoneal mass
(6 x 8 cm.) with widespread lymphadenopathy had been present for
3 months. Biopsy of cervical node revealed lymphosarcoma.

Day	Course
-6	Hgb. 8.8 gm; WBC 4,500
	Radiotherapy to abdomen (100r/1 day) 2 m v
-3	Patient very ill, bedridden, requiring heavy narcotics
	Bone marrow aspiration (320 cc. stored)
0	Total body irradiation 200r/7 days (100r x 2) 2 m v
11	Regression of all nodes and masses, jaundice cleared
28	Hematopoietic depression; Hgb. 6.8; WBC 866; Plts. 14,000
	Re-infusion of bone marrow
39	Continued depression; Hgb. 6.8; WBC 431; Plts 54,000
45	Beginning recovery; WBC 2,138; Plts. 90,000
68	Remarkable improvement; patient ambulatory
	Hgb. 11.2; WBC 3,900; Plts. 212,000
90	Remission continues; WBC 6,200
103	Recurrence of symptoms, developing lymphadenopathy
	Hgb. 12.1; WBC 2,880; Plts. 130,000
115	Radiotherapy to enlarging abdominal mass (150r/1 day)
	Moderate relief
151	Hgb. 8.1; WBC 1.710; Plts. 96,000. Expired.

Comment. Survival of 5 months post irradiation permitted obser-
vations throughout the depression and recovery period accompanied
by remission lasting nearly 3 months and eventual recurrence of
symptoms. During this time, multipe hematologic and biochemistry
studies were performed. In evaluating effect of radiation, the
marked anemia associated with the disease must be considered.
Severe anemia was present on admission and, although blood count
was normal following recovery from total body irradiation, anemia
recurred with exacerbation of symptoms. Thus, total body irradiation
was not solely responsible for the severe bone marrow depression
following treatment but probably was responsible for improvement
in the blood picture occurring 3 months post treatment by virtue of
improvement in general condition. Figure 2 shows response of
white blood cell count and total circulating granulocytes.

Among humans exposed to irradiation, the development of leukemia
(7, 8. 9), sterilization, and changes associated with premature aging have
been attributed to latent effect of irradiation, and practically all congenital
anomalies are consiered mutations arising from effect of radiation of
progenitors (10, 11). While genetic effects require generations to become
evident, and the amount of total body irradiation required for sterilization
far exceeds the levels used for therapeutic purposes, delayed effects
might be observed in patients with prolonged survival.

Fig. 2   Total Body Irradiation

Comparison of response of white blood cell count and
granulocytes following 200r/7 days

(J.C.)
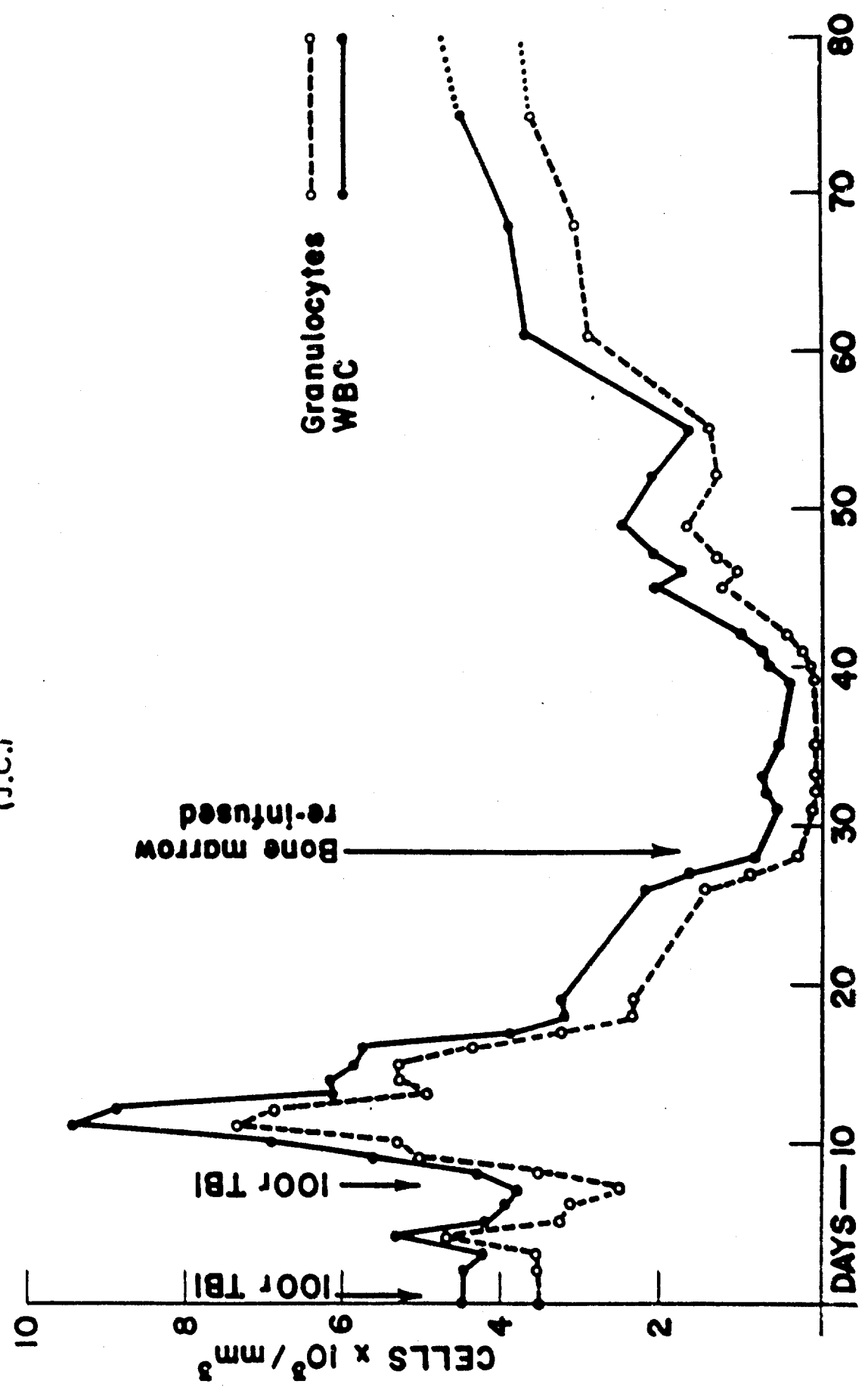
As with immediate radiation change, the reliability of evaluation
of latent effects must depend upon the accuracy of measurement of
irradiation, time lapse between exposure and observed change, and
measurement of change. In accidental exposure, or exposure associated
with occupatient (medical or industrial), the amount of radiation can
only be estimated while, in occupational exposure, both amount of
radiation and time lapse are difficult to ascertain. In any type of
exposure, latent changes due solely to irradiation are influenced by
mechanisms of continuous wear, aging and repair. In this series, 9
patients were followed from 6 to 10½ years and were observed for
possible late changes. These patients are particularly appropriate for
this study because amount of radiation is known and time lapse to last
observation has been established. Effect of radiation must be evaluated
on the basis of changes that: 1) are known to result from radiation,
2) are not compatible with age or disease, and 3) cannot be accounted
for by heredity or chance. Table 9 summarizes amount of radiation
exposure and present status with elapsed time for the 9 long-term
survivers.

Table 9

Patient	Age	Irradiation	Present Status & F.U. Blood Count
1. J.R.	83	545r/16 days	Died - HCVD 6 yrs. Post Rx. Age 89 Hgb. 13.5, WBC 9,000
2. S.P.	70	125r/1 day	Died - HCVD 7 yrs. Post Rx. Age 77 Hgb. 13.2, WBC 6,400
3. C.W.	64	75r/1 day	Lost - 7 yrs. Post Rx. Age 70 Hgb. 13.0, WBC 6,400
4. B.J.	27	200r/1 day	Lost - 7 yrs. Post Rx. Age 34 On parole from penitentiary, no final WBC
5. G.M.	24	50r/1 day	Living - 7½ yrs. Post Rx. Age 31 Hgb. 12.8, WBC 7,200
6. R.P.	28	25r/4 days	Living - 7½ yrs. Post Rx. Age 35 two children - ages 5 & 3 yrs. Hgb. 15.2, WBC 7,500
7. A.B.	54	50r/1 day	Living - 7½ yrs. Post Rx. Age 61 Hgb. 13.8, WBC 7,500
8. A.H.	60	50r/1 day	Living - 10½ yrs. Post Rx. Age 70 Hgb. 13.0, WBC 6,200
9. I.G.	75	100r/1 day	Living 10½ yrs. Post Rx. Age 86 Feeble, in nursing home Hgb. 10.0, WBC 4,400

The minimum amount of radiation causing specific delayed changes that can be noted clinically in humans cannot be determined; it may exceed the level of therapeutic exposures used, or more time may be required for change to become evident. In this group, no changes were noted that could be attributed to radiation exposure. J.R. (No. 1) received an exposure which, of the entire group, would be most likely to produce latent effect. Since the practice of radiotherapy must balance competing risks of disease with risks of treatment, this patient's age, history of bilateral orchiectomy and generalized disease were weighed against the possibility of radiation change. Tolerance was improved by p rotraction of radiation and there was excellent therapeutic response. The history of this patient warrants reporting.

J.R. This 83 year old man was admitted with signs and symptoms of a testicular tumor. Bilateral orchiectomy was carried out and pathology report was lymphoma. Six months later, he developed difficulty in swallowing and examination revealed a firm lesion on posterior tongue with enlarged right mandibular nodes. Biospy again revealed malignant lymphoma.

Day   Course
  0   Hgb. 14.1; WBC 8,000; Plts. 196,000
      Total body irradiation 545r/16 days
      Malaise toward end of course with some nausea & vomiting

 35   Mass at base of tongue decreased in size; mandibular nodes
      disappeared. Lowest point of hematopoietic depression
      Hgb. 12.8; WBC 3,000; Plts. 54,000

 67   Recovery well under way. Lesion of tongue disappeared.
      Hgb. 13.0; WBC 4,000; Plts. 117,900

355   Approx. 1 year post irradiation. Feeling well
      Hgb. 13.4; WBC 5,500; Plts. 180,000
      Normal activities

5 yrs. No complaints, no evidence of disease
      Hgb. 12.9; WBC 5,700; Plts. 170,000

2220 6 yrs. + 1 mo. Cerebral accident.
      Hgb. 13.5; WBC 9,000

2222 Expired.

Comment. Because of the likelihood of recurrent disease, this elderly patient was seen frequently for the first 2 or 3 years post irradiation and at 6 month intervals thereafter. Five years following treatment, he was normally active, required glasses only for reading, and had no complaints. Blood count 2 days before death was normal and there was no evidence of radiation change. He died following a cerebral accident at the age of 89 years. Comparison of response of granulocytes with total white blood cell count is shown in figure 3.

Total Body Irradiation

Fig. 3. (J.R.) Comparison of response of white blood cell count and granulocytes following 545r/16 days
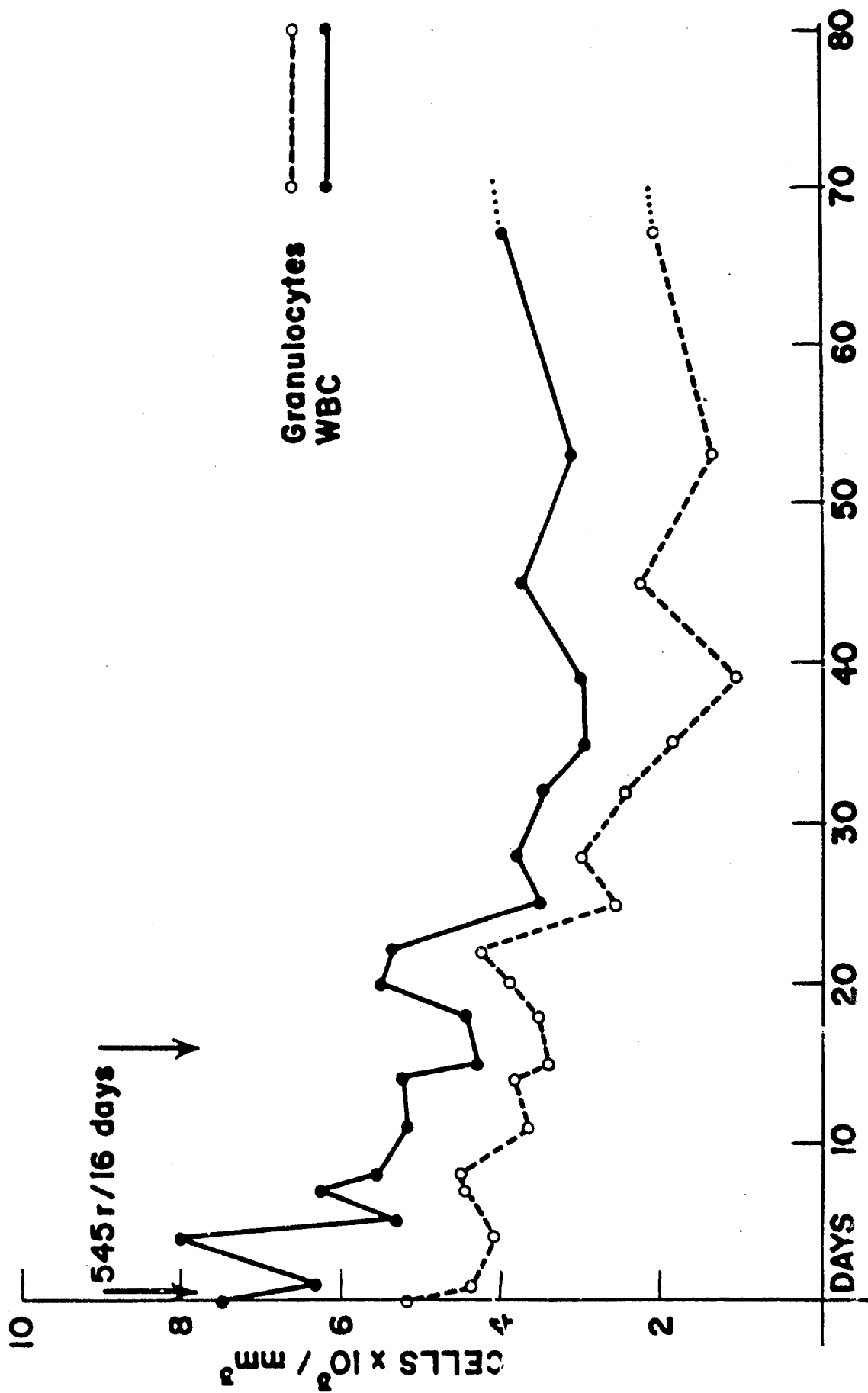
During the past few years, the protocol of procedures for patients under-going therapeutic total body irradiation was extended to include some 30 biochemistry tests, and physiology studies including gross body compostion and monitoring of vital functions. The protocol required that baseline studies be obtained prior to irradiation with all tests repeated at specific intervals following treatment. No conclusions on this aspect of the work could be drawn because: 1) a large number of patients would be required to provide a standard baseline for cancer patients and to observe changes that might be correlated with amount of radiation, 2) many patients were unable to tolerate multiple tests because of poor general condition, and 3) several patients died before the series could be completed. The complete protocol would have value if it could be applied to all patients receiving therapeutic total body irradiation in several centers throughout the country. This would provide an adequate number of patients for study with opportunity for proper evaluation. However, in the biochemistry studies, results indicate that several tests, namely urea nitrogen, glucose (fasting), serum amylase and serum lipase, merit further investigation.

In body composition studies (described in detail in the Progress Report for period ending 1/31/63), increase in extracellular water and decrease in intracellular water were thought to be due to stress of total body irradiation but changes in lean body weight and fat content could be attributed to weight loss associated with the course of the disease or to total body irradiation. The considerable variation in changes in blood volume led to an intensive study to develop suitable techniques for measuring blood volume and the application of these techniques to changes following other forms of stress.

Monitoring of vital functions with the 6-channel physiograph was carried out on several patients before, during and after irradiation. Constand recording of respiration (rate and depth), blood pressure, electro-cardiogram, electroencephalogram and cardiotach failed to reveal definit changes in function except for deceleration of heart rate which coincided with a feeling of nausea in two patients. Both of these patients received partial body irradiation, one to a large abdominal field and one to entire brain. The amount of irradiation to these areas exceeded the level used for therapeutic total body irradiation which may account for the changes noted. In animals, marked changes in vital function were noted following massive radiation exposures (see progress report submitted Jan. 31, 1963).

Animal Experiments

Studies in the latent and genetic effects of total body irradiation have been carried out in animals (usually rats, mice or rabbits) by means of biochemical tests and histologic examination of specific tissues (13, 14, 15, 16, 17). Some investigators have reported both histological and bio-chemical changes in specific tissues, particularly testis, liver and thymus (see Literature Review). These have contributed little information because:

1) experiments produce different results in the hands of different investigators, 2) in some experiments, the amount of radiation used to induce change causes death, preventing long term observations, 3) such changes are the direct effect of radiation and cannot be construed to reflect genetic effects if the animals had survived, 4) techniques of exposure in experimental work are apt to produce disproportionate absorbed dose, i.e., for a given amount of radiation the per cent fall-off in the middle of a small animal would be much less than for a large animal or for a human, and 5) the response of rats, mice and rabbits to irradiation differs considerably from that of man and the findings cannot be applied to humans. For these reasons, it is felt that studies on higher animals might be useful in determining effects that might be expected in humans, and all animals receiving sub-lethal exposures for various purposes throughout the study were retained for observation.

In the course of a study undertaken to investigate the role of bone marrow in recovery from heavy exposures of total body irradiation in dogs, a group of animals have survived from 9 months to 4 years. These animals provide opportunity for long term observations following moderate to heavy exposures ranging from 300r total body irradiation to 1200r total body irradiation with shielding of iliac marrow. In the latter group, four animals received 1000r to the previously protected iliac marrow. Tables 10 and 11 show the amount of irradiation, protection of iliac marrow, course, and survival time.

Table 10

Total Body Irradiation - Experimental Animals (Dogs) with Shielding of Iliac Marrow

No.	Sex	T.B.I. with shield	Iliac Marrow Irradiation	Survival	
57A	F	1000r	1000r $\bar{p}$ 9 mos.	3 yrs. 11 r s.	doing well
15A*	F	700r	none	4 yrs.	doing well
13A	F	1200r	1000r $\bar{p}$ 9 mos.	3 yrs. 11 mo:	doing well
9A	F	600r x 2/3 days	none	3 yrs. 4 mos.	doing well
16A	M	1200r with hydrocortisone	none	3 yrs. 3 mos.	paralysis hind quarters
31A	M	1000r	1000r $\bar{p}$ 14 days	3 yrs. 3 mos.	doing well
14A	M	1000r	none	3 yrs. 1 mo.	doing well
18A	F	1000r	1000r $\bar{p}$ 14 days	3 yrs.	doing well

*Animal bred 2 months post irradiation with unirradiated male; offspring (not included in table) 62A female, and 63A female, both normal.

Table 11

Total Body Irradiation - Experimental Animals (Dogs) with Bone Marrow Transplants

No.	Sex	T.B.I.	Bone Marrow Transplant	Survival
36B	M	300r	Autologous	9 mos. Doing well
47A	F	300r	none	9 mos. Doing well
35B	M	300r	Isologous	10 mos. Doing well
48A	F	400r	none	11 mos. Doing well

Moderate to severe bone marrow depression developed in all surviving animals but, with recovery, all blood counts returned to normal. Except for one dog (No. 16A), all animals are healthy and frisky; there is no evidence of cataract or other damage to lens, and no evidence of anemia or other blood dyscrasia. No. 16A developed paralysis of hind-quarter 2½ years post irradiation. X-rays reveal no abnormality and the animal's blood count is normal; further investigation is being undertaken. In appearance, animals with light-colored coats are unchanged. One black-coated dog is completely gray and the black area of one black and white dog is gray.

One animal, not included in the tables, received 1400r total body irradiation with iliac marrow shielded. This dog recovered but died of distemper 37 days post irradiation. At the time of death, there was no clinical evidence of radiation injury; his white blood cell count was 8,150 and platelets were 70,000. Autopsy showed normal activity of marrow of iliac crest with many mitoses in marrow of long bones.

In this laboratory, proper preparation of animals and studies in dosimetry to deliver an even distribution of radiation have made it possible to establish the LD-100 for dogs at 700r total body irradiation. One animal (15A), receiving 700r with shielding of iliac marrow was bred two months post irradiation. This dog has now survived 4 years. The offspring (62A and 63A) were healthy, normal puppies, developed normally and are now adult animals. If these first generation post irradiation animals could be maintained and bred with other animals surviving moderately heavy exposures with and without shields, and with unirradiated controls, they would represent the origin of possible mutations in future generations. A breeding schedule, designed to demonstrate transferable effects of radiation, has been prepared but it is recognized that such a project is impractical because of the many generations required to produce observable genetic effects.

Apart from the possible origin of genetic effects, continued observation on all surviving animals for latent radiation effects is of considerable

interest. If the amount of radiation exposure received by these animals is sufficient to cause delayed effects, then the development of anemia, leukemia or other malignant process, and acceleration of the aging process (other than graying coat) might be expected. Since the time for delayed effect to become evident varies (7, 8, 9, 13), only continued observation offers the possibility of relating amount of radiation, time and radiation change. A review of reports of animal experiments dealing with total body irradiation by other investigators indicates that no other series receiving near lethal exposures has been maintained long enough to make these observations.

Granulocyte Proliferation Study

The peripheral blood count has always been considered the best index of radiation change but the nature and shape of dose response curves of major classes of cells do not fully explain the role of bone marrow in recovery from total body irradiation. Bone marrow depression is reflected in peripheral blood cells originating in bone marrow, i.e., red blood cells, platelets and granulocytes. The time at which changes in these elements will become evident is a function of the rate of production and the survival time. Granulocytes, having the shortest life span (up to 4 days), are the first to reflect radiation change. Among animals receiving lethal exposures of total body irradiation with a portion of iliac marrow shielded, it was possible to study production of granulocytes by bone marrow examinations and by peripheral blood counts.

The compartments or stages of development of the precursor granu-locytic elements found in bone marrow have been described in terms of number of cells and time spent in each compartment (18, 19, 20). Using information obtained from the literature and observations from our own experiments, a model of granulocyte proliferation was constructed for humans and for dogs. The proliferation pathway for each is the same excep: that time spend in each stage of development in the dog is shorter than that in man.

This is a self-perpetuating system at the stem cell level. As the result of a single division in a 48 hour period, two daughter cells are produced. One retains the characteristics of the parent cells and divides again in 48 hours; the other goes through transformation to a myeloblast. This is followed by successive divisions in promyelocyte and generative myelocyte stages with transformation through the other stages. A single stem cell will release 16 mature segmented neutrophils into circulation every other day. There is another group which releases mature segmented cells into circulation on alternate days. In the steady state, self-perpetuation is the result of self duplication at the stem celll level. To obtain an in-crease in stem cells proportional to the rate of recovery following total body irradiation, these cells need only to reduplicate one additional time. The result is 4 stem cells, two return through division the same as the

original parent stem cell of the steady state cycle. The result of this altered stem cell cycle is the creation of a steady state cycle every other day. As each myeloblast results in 8 mature granulocytes in circulation. the ratio of cells being added to the peripheral circulation can be determined.

Utilizing this modification, the recovery rate in terms of circulating granulocytes can be determined with indications to the apparent stage of development in marrow at the time of irradiation. Calculations indicate that the number of cells in each compartment of development surviving the direct effect of radiation can be evaluated quantitatively and correlated with amount of radiation.

In the application of this aspect of granulocyte proliferation to humans receiving therapeutic total body irradiation, a quantitative estimate of cells may be compared to the observed peripheral granulocyte count. In a patient receiving 200r therapeutic total body irradiation, comparison led to the following conclusions.
1. Cells present in peripheral circulation at the time of exposure are not destroyed by this amount of radiation; survival time in circulation is equal to the pre-irradiation value.
2. Cells which are in the transformation compartment continue on to maturigy.
3. All cells in the dividing-transformation stages are lost.
4. Approximately 20% of stem cells in the pre-myeloblast stage are lost; the remainder continue to normal maturity.
5. 50% of parent stem cells are lost; 7 % are altered to form the non-steady state recovery cycle. The remaining 43% continue division according to the steady state cycle but only until the 11th or 12th day. These cells are apparently unable to continue division and are lost.
6. Only those cells produced by the altered cycle are capable of continued division, leading to eventual recovery.

A quantitative analysis of the number of stem cells destroyed by irradiation can be determined by the 14th day post irradiation. Plotting the number of destroyed stem cells against increasing air dose, the resulting sigmoid curve closely resembles the LD values determined for dogs and postulated for humans by Cronkite (21). By extrapolation from estimates obtained by work on experimental animals, effect of higher doses of radiation in humans may be estimated. In both animals and humans, calculations indicate that repopulation of depleted marrow can be accomplished if as little as 6% of stem cells survive. A comparison of granulocyte count with total white blood cell count following total body irradiation is shown in figures 1, 2, and 3.

Studies in granulocyte proliferation have particular application in bone marrow replacement for depression following total body irradiation. If each transplanted stem cell is established in marrow sites on the day of infusion and maintains its ability to divide and progress through the steps of maturation, progeny cells would appear in circulation about the 10th day post infusion. Following single exposures of therapeutic total body

irradiation, the time of greatest depression occurs 25 to 30 days post treatment. Thus, if calculations are correct and bone marrow replacement is an effective treatment, severe bone marrow depression might be avoided if transplantation is carried out prior to the 10th day post irradiation.

Work so far indicates that cells of the granulocyte series may offer advantages as a biologic index of radiation and, by comparison, may permit estimation of amount of irradiation received, as in cases of accidental exposure. The study also provides a basis for estimating: 1) the amount of radiation that would cause death by bone marrow depression, 2) the amount of marrow that must be protected or transplanted to sustain life following exposure and 3) the time when transplanted marrow would be of greatest benefit.

Bone Marrow Bank

Developed first in animals, techniques of aspiration, processing, storage and re-infusion of bone marrow were quickly extended to humans. Since that time there has been no lack of clinical material for bone marrow studies. Marrow is aspirated and stored at -80° C. for all patients receiving therapeutic total body irradiation, for many patients receiving chemotherapy, and it has been recommended that patients in remission from disease have marrow stored against the time when exacerbation of symptoms causes severe blood dyscrasia. Autologous bone marrow has been re-infused in humans, and autologous and homologous marrow has been transplanted in animals.

Samples of marrow are tested for viability at the time of aspiration, at intervals during storage, and prior to re-infusion. In some instances, the amount of marrow obtained was small and the entire amount has been used for repeated viability tests. Among the patients whose marrow is presently in storage, the majority have died but marrow is saved to check decreasing viability with increasing time in storage. Table 13 summarizes the activities and present status of the bone marrow bank.

Table 13
Bone Marrow Bank

Patients

Marrow aspirated, processed and stored		70
Marrow re-infused	10	
All stored marrow used for viability studies	16	
Marrow now in storage	44	

Animals

Marrow aspirated, processed and stored		72
Marrow re-infused or transplanted	62	
Marrow in storage	10	

Using standard tests, viability of marrow at the time of re-infusion has been approximately 90% but, clinically, this has been difficult to con-firm. Among the patients and animals studied, marrow cells surviving the direct effect of radiation may be responsible for recovery and, except in cases where exposure is of the order of LD-100, survival itself is not proof of viability of re-constituted marrow. Presently, there is no satis-factory way of determining viability and, if viability were certain prior to re-infusion, there are still problems of determining whether viable marrow cells find their way to marrow cavities and proliferate, and whether pro-liferation is sustained to contribute to recovery of bone marrow depression. Experiments have been designed to overcome these difficulties by in-vivo labeling of marrow cells with tritiated thymidine in experimental animals.

Observations on the effect of total body irradiation in humans and animals have given rise to problems require basic investigation and each year experiments have been conducted to provide the needed information. These have dealt with the role of total body irradiation in suppressing immunologic response to transplants of organs and tissues (32, 24.) and with experiments designed to confirm the work of other investigators. During the past year, an experiment was undertaken to determine lethality in mice following total body irradiation; results are summarized in Table 14.

Table 14
Lethality of Mice Following Total Body Irradiation (220 Kv.)

Exposure	650r	700r	750r	800r
No. of animals	27	30	30	50
LD-50	15 days	10 days	8 days	7 days
LD-65	20½ days	16½ days	9 days	8 days
LD-75	0*	23 days	11½ days	8½ days
LD-100	0*	0**	14 days	13 days

*35% survivals; ** 25% survivals.

The surviving animals have been followed for more than 8 weeks with no further deaths.

Although it is recognized that lethality curves for mice (or rats) cannot be extrapolated to humans, experiments with small animals demonstrating good correlation between amount of radiation and response are deceptively simple. In 1950 Hennessy and Huff reported distinct correlation between radiation exposures ranging from 5r to 25r and erythrocyte depression as determined by iron turnover studies in rats (22). The experiment was repeated in this laboratory, using identical methods, materials and exposures but no correlation between dose and response could be established. In both clinical work and research dealing with total body radiation, experience

indicates that exposures of 5r to 25r are not likely to produce changes in the hematopoietic system that can be detected by any parameter available for measurement.

Literature Review and Bibliography

Because of the apparent gap between basic investigation and clinical observations on total body irradiation, a review of articles and reports dealing with this subject was undertaken. The material, covering a 15 year period (1947 - 1962), necessarily included related publications concerning detection, dosimetry, hazards and protection. Among those dealing with protection, a considerable number were devoted to anti-radiation drugs.

Of more than 600 reports, 78 were selected because: 1) they were representative of all studies concerned with biologic and physiologic response to radiation, 2) results were recorded, 3) results were supported or contradicted by different investigators. In the entire survey, only 10 reports concerned total body irradiation in humans and these are included in the 78 selected. Reports concerning accidental exposure were omitted because amount of exposure was uncertain. Among the 10 concerned with humans, four dealt with continuous low-level exposures of medical and industrial personnel (presumably exposures did not exceed the maximum permissible dose). One (Cronkite - No. 22) discussed clinical response following heavy accidental exposure (i.e., nausea, vomiting, loss of consciousness, etc), three dealt with studies in renal function, one with renal and liver function and one (Nickson - No. 59) emphasized hematology with one of two patients showing electroencephalogram changes following irradiation to the head. Reports on individual cases of accidental exposure were omitted because amount of radiation was uncertain.

Since work in this laboratory on humans has provided ample information on response of the hematopoietic system to irradiation, no effort was made to compile similar data on small animals. No reports were available on the possible application of granulocyte response as a biologic index of radiation exposure.

The 78 reports are listed with name of investigator, reference, amount of irradiation, species and results (pages 21-33)   and results have been summarized according to organ or tissue studied, type of test, amount of radiation, species and reference (pages 34-37).   A diagram has been prepared showing studies designed to demonstrate changes at the molecular level, biologic effects, metabolic disturbances, morphology, and recovery or death (figure 4).

Among the 68 reports representing many types of gross and histologic examinations and biochemistry studies in animals, in vivo and in vitro, results vary considerable, with frank contradiction in some cases (see increase vs. decrease of weight of testis, Nos. 9, 3, 77, 78; increase vs. decrease in survival with increasing energy Nos. 26 and 31). Variations

may be due to differences in species, size of animals, amount and quality of irradiation, uneven distribution of radiation, and single or protracted exposures. Approximatly one-half of the investigations were carried out on rats and mice while the remainder included rabbits, guinea pigs, ducks chickens, dogs, monkeys and in vitro studies.

Since it is not possible to investigate changes due to moderately heavy radiation exposures in normal humans, investigators must study animals with a view to comparing and extrapolating response to that which might be expected in humans. However, the value of animal experiments for this purpose is questionable when exposures exceed the LD-100 for humans. Following exposures of 10,000r, 30,000r or 750,000r, it seems likely that any looked-for change could be noted and that any stress of comparable severity, such as boiling in oil, would be equally effective. In the maze of incomparable material, comparison of results is not possible and extrapolation of results to that which might be expected in humans would not be meaningful. Yet, the survey has value in pointing out the many approaches to the problem while indicating the areas of need. These areas include:

1. Despite all the data available on hematologic studies, a definite quantitative relationship between amount of radiation and response has not been recognized.

2. Information concerning response to irradiation in humans by means of biochemistry studies is incomplete.

3. There are no reports dealing with physiological studies of total body composition and vital function changes following irradiation.

4. The contribution to the effect of total body irradiation of other stress or injury that might be encountered has not been explored.

5. Results of numerous, independent investigations cannot be evaluated because of diversity of materials, methods and aims.

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & Am't. of radiation	Findings
1.	Albaum, J. Serum enzymes after whole body radiation. Rad. Res. 12:186-194, Feb. 1960	rabits 700	Of 14 enzymes studied, glutamic oxalacetic transaminase was best early indicator of damage. 3 enzymes decreased, 4 were elevated. Sham studies caused changes in 10 of 14 studied.
2.	Allegretti, et al. Effect of whole body irradiation on Langerhans islets. Rad. Res. 13:31-36, July 1960	guinea pigs 250r	Histological examination of beta-alpha cell ratio; smallest islets showed greatest increase at 24 hrs. with later shifting to larger islets. Transformation of islet cells can occur.
3.	Allen, J.D. & Jacobson, L.O. Hyper-heparinemia: cause of hemorrhagic syndrome p total body irradiation. Science 105:388, 1947.	Dogs ? dose	Thrombocytopenia plays only secondary role. 1. Radiated dog blood delays clotting of normal blood. 2. Antiheparin substances returns clotting time to normal in vivo & in vitro. 3. Isolated heparin-like compound 4. No correlation between hemorrhage & thrombo-cytopenia 5. Vit. K, ascorbic, Ca. salts & whole fresh blood transfusions did not prevent onset of hemorrhage
4.	Alpen, E.L, et al. Effect of radiation on lymphoid tissue in vivo. Intern. J. Rad. Biol. 2:425-439, 10/60 NSA 1/31/61, p. 169	rats ? dose	DNA synthesis of large & med. lymphocytes were more radiosensitive in vivo than in vitro. Histological & autoradiographic techniques permit separation of effects due to population changes from those due to biochemical alteration. Among controls, average generation time for large & med. lymphocytes of lymph nodes is 20-30 hrs. Evidence is in favor of a longer life span for the small lymphocytes.

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & Dose	Findings
5.	Anderson, D.R. Effect of radiation on creatine metabolism. Rad. Res. 1957, p. 300	1500r rats	Marked creatinuria (20 x normal). 3 - 4 fold increase in plasma creatine. No change in urine creatinine or or guanido-acetic acid.
6.	Barron, E.S., et al. Mechanism of action of ionizing radiation. I. Inhibition of enzymes. J. Gen. Physiology 32:537-552, 1949	in vitro 25 - 500r	Inhibition of phosphoglyceraldehyde dehydrogenase 50% after 200r, 100% after 500r. Hexokinase - little change. ATP at pHG.1 inhibited at 10r with 100% at 1000r. Succinoxidase inhibited proportional to concentration. No inhibition of enzymes without sulphydral groups.
7.	Ibid. Idem. 32:595-605, 1949 II. Inhibition of sulhydral enzymes		Inhibition was dependent on type of radiation (alpha, beta, gamma) with some effects at 14r. Maximum occurred by the 6th day.
8.	Bettendorf, G. Berlin. Strahlenterapic, Urbon & Schwargenberg 112:74-78, 5/60 Effect of total body radiation in intermediate produces of nuclei acid metabolism.	rats 800r	Changes in activity & concentration studied by labeled phosphate. Those showing decrease in thymus by 30 min. were: ADP, GTP, TTP, DPN+ changes at 7 hrs. which were: AMP, ATP, CTP, UTP. Decreased activity shown by P32 uptake by 30 min. in TTP, AMP; by 7 hrs. in ATP, CTP, GTP, UTP, & RNA.
9.	Boling, J.L. & Floyd, R.L. Biological effects of high intensity microsecond pulses of x-irradiation ac determined by post-irradiation testicular weight loss. DA-49-007-901, Report 3/60. TAB 2/1/62, p. 104.	mice	Testicular weight loss doe to irradiation at high dose rate is roughly comparable with that reported due to relatively low dose rate.
10.	Bond, Fliedner & Cronkite J. Nuc. Med. Oct. 1960		Results summarizedseparately
11.	Botkin, et al. Thyroid response to total body irradiation. Endocrinology 50:550, 1952	rats 1000r	Increase in thyroid activity after 2 hrs. apparent for 24 hrs. Normal by 6th day.

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
12.	Bowers, J.Z. & Scott, K.G. Distribution & excretion of electrolytes p acute total body irradiation injury. Studies in radio-sodium Proc. Soc. Exp. Biol. & Med. 78, 1951	rats 710r	Edema when histopathological findings were pronounced. Occurred when radio Na & K were decreasing. Decrease in urinary excretion of radiosodium corresponds with blood decrease and increased G.I. injury.
13.	Bowers et al. Effect of acute whole body irradiation on salt & water metabolism & significance. Radiology 61:97, 1953.	rats 660r	Decreased food & water intake. Negative balance of Na, K, Cl for 1st 3 days. Loss of K & gain of Na in small intestine at 24 hrs. Increase of K in liver @ 24 hrs.
14.	Brent, R.L. et al. Effect of radiation on serum transaminase level. Rad. Res. 1957, p. 305	700r rabbits	Increase in glutamic acid oxalacetic transaminase within 24 hrs. No correlation to pre-irradiation values or death.
15.	Breyer, F.T. et al. Effect of radiation on adreanl cortical steroid excretion in urine. Science 120:112, 1954.	pigs 750r - 1000r	Marked increase in excretion of adrenocortical steroids within 1st 24 hrs.
16.	Brown, C.S. et al. Biochemical, cellular & bacteriologic hanges in thoracic duct lymph p total body irradiation. Am. J. Physiol. 163; 668; 1950	dogs 500r	Decrease in T. protein within 50 hrs. Increase in NPN in 5 hrs. Decrease in keratin values by 30% by 17 hrs. Sl. change in sugar. No change in Cl. Lymphs decreased to 18% by 18th hr. with low of 13% by 132 hrs. These results slightly different from blood studies done at same time. Bacterial cultures negative on blood & lymph.
17.	Brown, S.O. & Krise, G.M. Chronic whole body radiation stress. Ann. Prog. Report 2/61. Radiation Biol. Lab. College Station	rats & mice 0,2,5,10,20r daily	Decrease - WBC, platlets, erythroid elements @ 10r/day No change in glucose homeostasis. Decrease in tolerance to therman stress.
18.	Carter, W. Variation in tissue DNA p total body irradiation. Fed. Proc. 12:188, 1953	rats, rabbits LD-50	Tissues: muscle, skin, gut, liver, kidney, spleen & testis Analysis: DNA, H_2O, fat, Na, K, Cl. Marked increase of DNA in all tissues except kidney during 1st 4 hrs. In rats - decrease in intra- extracellular fluids

Effect of Radiation - Biochemistry Studies

	Title & Reference	Organism & dose	Findings
19.	Cavalieri, R.R. et al. US Naval Hosp. Decreased urinary excretion of 2-aminoethanol by irradiated humans. NSA 4129 Nature 191:1303-1304, 9/23/61	humans 300r-900r	Among normal controls & cancer patients, urinary excretion of aminoethanol, although variable, was never less than 10 mg./day; p whole body irradiation, aminoethanol decreased to undetectable concentrations (less than 0/4μ gm/ml., reappears by the 12th day.
20.	Conrad, R.A., Cronkite, E.P. et al. 3/56 Experimental Rx of G.I. tract syndrome produced by lethal doses. Naval Med. Research Inst. Bethesda	Dogs - local lethal	Anorexia, vomiting, diarrhea, fluid & electrolytes loss and vascular collapse.
21.	Constant, M.A. & Phillips, P.H. Effect of multiple low-dose radiation & splenectomy on stability of dog erythrocytes. Am. J. Physiol. 178:367, 1954.	dogs 100r/wk.	Marked decrease in fragility when compared to non-splenectomized, irradiated dogs. Both were lower than normal (less fragility). Rise noted in several in terminal stage. No correlation to life expectancy, of Hgb. levels, platelets or target cells.
22.	Cronkite, E.P., & Bone, V.P. Diagnosis of radiation injury & analysis of human lethal dose of radiation. Armed Forces Med. J. March 1960	humans dose-variable	Human data collected from all accidental exposures see summary (No. 10)
23.	Detrick, L.E. et al. Thiamine transport across irradiated rat intestine Rad. Res. 15:520-526, Oct. 1961	rats 525r TBI	Following 525r there is decreased absorption of thiamine probably related to subcellular damage in crypt cells in rats.
24.	Dicksen, E.A. & Shapiro, B. Effect of gamma radiation on viscosity & enzymatic activity of urease solution. Rept. No.61-85, Sept. 1961, TAB 2/1/61, p. 101	in vitro Co-60 750,000r	Specific viscosity of urease decreases initially, then levels off at about 3/4 the original value. Protection against irradiation produces viscosity in urease solutions containing (GES)2 which inhibits enzymatic activity, probably due to intramolecular rearrangement
25.	Dorfinon, R.I., Rosenfeld, G. Steroid excretion in calf urine. 1/58 Naval Med. Res. Inst. Bethesda	calf	Urine 17-keto. & 17-hydorx. - increase utilization of I.V. hydrocortisone

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
26.	Ellinger, F. et al. Lethal effect of 200 & 2000 Kv. x-rays & Co-60 gamma rays. Rad. Res. 1957, p. 313.	guinea pigs 325-750r	LD-50 dose increased by 1/3 to ½ between 200 Kv. 2000 Kv and Co-60. Definite dependency on type of radiation & exposure geometry on mortality.
27.	Esnouf, M.P. et al. Biochemical studies 6 - 18 mos. post TBI. J. Radiat. Biol. 3:459-466, 9/61	mice	There was reduction in the incorporation of P^{32} into kidney DNA relative to that in RNA
28.	Ibid, idem p. 467-473, 9/61	mice	Irradiation produced permanent depression of DNA synethesis relative to RNA in the kidneys
29.	Fransen, Fr. Ann. Tech. Rept. Cologne, U. Germany. Formation & pathophysiological significance of biogenic amines ñ sublethal radiation injuries.	rats 500r	3-hydroxytyramine - results not reported noradrenaline & histamine - results not reported 5-hydroxytryptamine - urine increase 6th-10th day 5-hydroxindol - increase 2 x in urine Ethylamine & isoamylamine - no increase Allylamine & vinylamine - both increased
30.	Geisselroder, J. & Nims, F.F. Carbo-hydrate metabolism in stress. Rad. Res. 1957, p. 317	rats	Blood sugar, liver glycogen and muscle glycogen showed increases
31.	Goldfeder, A. & Clark, G. Evaluation of "Time Factor" in radiobiological effects Rad. Res. 1957, p. 318.	mice 700r	700r @ 8.6r/min, 25% survival 700r @ 17.2r/min. 47.8% survival 700r @ 830r/min, 66.6% survival. No significant weight loss of whole body, spleen, liver or testis.
32.	Graham, Douglas & Sacher. Chronic radiation mortality p single T.B.I. Rad. Res. 1957, p. 319.	mice	Increasing LD-50 with decreasing energy (250 Kv-634r; 135 Kv-663r; 80 Kv-816r) no sex differences, no leukemia, tumors or degenerative disease differential for all qualities. Same relative effect.

Effect of Radiation - Biochemistry Studies

	Title & Reference	Organism & dose	Findings
33.	Hanford, S.W. Acute radiation syndrome p TBI in supralethal dose 8/19/60, Vol. 18 (report) Naval Med. Res. Inst. Bethesda	dogs LD-100/88hrs. Co-60	C.B.C. - decrease, all elements. Ser. Prot. & electrolytes - no changes Blood vol. - significant decrease at pre-mortal period Hepatic blood flow & renal blood flow well maintained until immediate premortal stage.
34.	Hale, W.M. & Stones, R.D. Effect of ionizing radiation on immunity. Radiation Res. 1:459-469, 1954		Data from various investigators: inhibitory effect on antibody response is intimately related to dose, species, time of stimulus, soluble or particulate nature of antigen, root of stimulus. Radiation reduces or abolishes active or passive immunity to bacterial & animal parasite infection. Little or no effect on acquired immunity to viral infections or bacterial toxin.
35.	Hawrylewicz, E.J. Effect of TBI on blood enzymes. 6/30/60 Armour Research Foundation, Chi.	rats 0.3-300r/wk. for 26 wks.	Co-enzyme A, aldolase, Glu-6-phosphate dehy.. Glu. Oxol. Trans, DPN - all increased with 300r/d - 300r/wk. Catalase LDH, carbonic anhydrase, cocarboxylase, flavin adenine dinucleotide - none showed any specific change.
36.	Ingram, M., et al. Occurrence of lymphocytes with bi-lobed nuclei in cyclotron personnel. Science 116:706, 1952.	humans low level continuous	Increase in lymphs with bilobed nuclei Returned to normal after protection
37.	Jackson, K.L. et al. Electrolyte excretion following severe intestinal damage by x-irradiation. Naval Radiolog. Defense Lab. San Francisco 6/57	rats 1500r	27% increase in K excretion in urine over 4 day interval 138% increase in sodium (in G.I. tract)
38.	Jacobson & Krohn, D.O. RBE of neutrons by correlating physiologic change of eye Cont. AF33(616)712, proj.7165 TAB 2/1/62, P. 105	rabbits	Eye damage based on electroretinogram, biochemical determinations, histopathological & clinical observations @ 24 hrs. post exposure. RBE varied with different indicators but appeared to be not less than 1 & not greater than 3. Higher energies appeared to have much greater RBE.

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
39.	Kaufman, J. The coagulogram as a critical indicator of irradiation effect. Am. J. Roentgenol. 55:464, 1946	humans low level	Platelets, clot retraction, bleeding time, fibrin time, complete coagulation time & prothrombin time altered fairly consistently.
40.	King, E.R. Use of total body irradiation in Rx. of far advanced malignancies JAMA 9:10, 9/2/60	humans 300r-500r	No increase of taurine urinary excretion until 300r or over. B-aminoisobutyric acid increases. Fibrin deposit in lungs after large single doses.
41.	Kinsey, V.E. Effect of neutrons & other radiations on ocular lens. Contr. AT(11-1) 152. Kresge Eye Inst. Detroit NSA 12/15/61 p.3925	animals	Study concerns the transport of amino acids across the blood aqueous barriers, the capsul & epithelium of the lens by relative concentration of amino acids in aqueous & vitreous humors, lens & plasma. No results so far.
42.	Klein, J.R. Brookhaven. Protoporphyrin & heme formation by erythrocytes from irradiation & bled duck Am. J. Physiol. 210:663, 10/7/61 NSA p.4128	ducks 800-1200r	In vitro studies show an increase in pro heme & free protoporphyrin formation by erythrocytes p frequent bleeding. Decrease in heme formation but not in free prophyrin production was observed.
43.	Klouwen, H.M. Radiosensitivity of nuclear RNA. Biochemica et Biophysics Acta 12:366-368, 8/12/60	rats 700r	In separated nuclei from irradiated liver & thymus cells, RNA was labeled with P^{32}; liver cells showed slight increase while thymus cells showed marked decrease in uptake.
44.	Kohn, H.I. Changes in blood plasma during acute radiation syndrome. Am. J. Physiol. 162:703, 1950	rats 200r	Glucose increased 60 mg.% 1st day, normal 5th day. NPH elevated for 5 days; A/G ratio rose from 3.1 to 4.5 1st 2 days, normal 5th day, due to nonprotein fractions. Decreased total protein 5th-7th day. Elevation chloride & cholesterol 3rd-5th day.
45.	Krise, G.M., et al. Radiation creatinuria School of Aviation Med. Randolph AFB Oct. 1957	rats 435 rep neutron-gamma	p mixed irradiation, no change p 5th day in urine creatine; with gamma only, creatinine decreased and creatine increased.
46.	Idem, Ibid May 1957	750r gamma 1500r gamma	1500r - 20 to 30 x creatinuria on 2nd day; 435r mixed - 36 to 63 x creatinuria on 2nd day; profound creatinuria (85 x) at death.

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
47.	Kawade, M. Nippon. Effect of x-rays on the affinity of liver for protoporphyrin III. NSA 8/31/61, p. 2666	guinea pigs 100r TBI	Slight change in parenchymal cells with some decrease of affinity for protoporphyrin, but after 14 days, metabolism returned to normal. p 400r-1000r, there was degeneration of parenchymal cells but no change in interstitial tissue & epithelial cells of bile ducts.
48.	Ibid, idem	local 1000r humans 2000r guinea pigs	2000r hepatic radiation strongly impaired dye excretory function of liver by BSP test. Administration of B_2, B_{12}, or glucuronic acid each day p exposure - disturbance was reduced. BSP in pts. receiving 1000r over upper abdomen showed similar results.
49.	Ibid, idem Re: renal function	Guinea pigs 50r-8,000r	Renal function decreased p exposure. Regardless of dose, quantity of urine was decreased; P^{32} in bladder was decreased in accordance with dose while P^{32} in blood & renal area was increased.
50.	Lamberts, H.B. An immediate radio-biological effect which is simple to record. Medica Mundi. 6:44-47, 1960	cock's comb, rat & human skin	Changes in macromolecular mucopolysaccharides. There was immediate reaction giving a drop in viscosity of synovia as measured by pressure of injection.
51.	E.H. Labrosse & Smith, J.E. Urinary coproporphyrin excretion p gamma Irradiation. School of Aviation Med. Randolph AFB 2/56	rats, 100r, 250r, 500r, 750r & 1000r	No dose relationship with coproporphyrin in urine
52.	Leone, C.A. Effect of irradiation as revealed by serological analysis. Tr. Kan. Acad. Sci. 60:301-315, 1957	chickens 330-990r	330r - no changes; 660r depressed activity at 24 hrs, normal p 3 days; at 30-60 days, serological activity exceeded that of controls. Major changes occur in the B globulins of the plasma
53.	Lott, J.R. & Wilding, J.L. Attempt to demonstrate lethal toxin in blood p irradiation. Sch. Aviation Med. Randolph AFB, 6/56	rats 1000r - 20,000r	No toxin demonstrated on gross transfusion

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
54.	Luzzio, J. Electrophoretic patterns c x-irradiated blood serums. Army Med. Res. Lab. Fort Knox. Report AMRL Proj. 6 X 64-14-001-01	In vitro	Electrophoretic changes produced in vitro irradiation of serum with massive doses parallel those changes observed after in-vivo exposure to lethal and sub-lethal exposures.
55.	McGoodall & Long. Effect of whole body radiation on adrenal medulla & on adrenalin & noradrenalin. Am. J. Physiol.197:1265-1270, 1959	rats, cats rabbits 800r - 30,000 r	Little change in noradrenalin titre p 800r but immediate decrease in adrenalin which returned to normal by 64th day; at 1600r decrease to zero; return of synthesis noted by 72 hrs. Rabbits & cats supported rat study.
56.	Milch, LJ. et al. Plasma lipoproteins & muscle adenosonetriphosphate in rabbits p local radiation. Sch. Aviation Med. Randolph AFB. Dec. 1955	rabbits 30,000r to hind legs	Varying dose rates. Increase in plasma lipids, decrease in lipoprotein, independent of dose. No change in muscle ATP
57.	Mole, R.H. Whole body irradiation & the idea of stress. Brit. J. Exper. Path.37:528-531, 1956	rats 200r	Release of cell contents from destroyed cells is responsible for the increase in blood sugar p irradiation, not increased adrenal activity. Increased liver glycogen is not due to increased adrenal activity.
58.	Nickson, JJ. & Koch, H.J. Postirradiation syndrome in humans. Coproporphyrin excretion, serum protein levels. Report #3 AFSWP-743, 3/31/55	humans	Results not reported
59.	Ibid, idem. Dec. 1959	humans 25rx5/5days 88r-130r/ 1 day	Detailed biochem. studies; hematology gave most consistent results. Of 2 pts. receiving irradiation to head, + eeg., 1 recorded subclinical explosive phenomena at 4 and 6 hrs.
60.	Parizek, J. Excretion of desoxycytidine in urine p irradiation. "The Cell Nucleus" N.Y. Academic Press, N.Y. 1960, p. 82-88. NSA 12/15/61, p. 3918	rats	Irradiation increase the excretion of desoxycytidine. Amount correlates well with amount of radiation even at relatively low dose levels (dose not given).

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
61.	Pitcock, J.A. Late testicular lesions in irradiated monkeys (path. & clinical studies) Report # 61-72. TAB 2/1/62, p. 101	monkeys 57 rep- 154 rep	Testicular lesions were found 3 - 8 years p exposure to gamma radiation. Degree of damage was independent of dose rate & fractionation bu is dependent primarily on the cumulative dose.
62.	Roth, L.J. Irradiation effects on C.N.S. Contract AT(11-)-847 NSA 3925, 12/15/61	rats 10,000r	No significant change in permeability properties of the blood brain barrier was detected for the human serum albumin or sulfate in irradiated brain.
63.	Sandberg, A. & Doull, J. Influence of low level gamma & fast neutron irradiation. I. Effect of aging & irradiation on adaptation to decreased barometirc pressure & glucose tolerance. A.F. Radiation Lab. Chicago. NSA 12/15/61, p. 3924	mice	Aged mice exhibit impaired glucose utilization, and irradiation exposure in the sublethal range decreases ability of both young & old animals to eliminate excess blood glucose.
64.	Seston, N., et al. Effect of sublethal TBI on glucose tolerance. Rad. Res. 13:25-30, 1960	rats guinea pigs 250r, 500r	Greatest impairment of glucose utilization on 8th day Believed to be extra-pancreatic in origin.
65.	Shiels, D.O. Structural aspects of lympho-cytes & monocytes relative to clinical condition p irradiation. Brit. Med. J. 32:306.	humans low level	Increase in large lymphs with decrease in lymphs showing granules. Increase in monocytes of a particular type. Significant changes in ratios of these cells related to exposure.
66.	Skalka, M. (Czech) Transaminase level in blood plasma p single & repeated doses. NSA 12/15/61, p. 3927.	mice	Increase in glutamic-oxalacetic transaminase (GOT) & glutamic-pyruvic transaminase (GPT) in plasma. Max. increase @ 8 hrs., a second peak may occur @ 7 days. Further increase when irradiation was repeated p 4 days but repeat exposures after 7 or 10 days produced no increase.
67.	Slenson, J.R. Effect of TBI on some con-constituents of liver & kidney. Proc. SocExper. Biol.&M. 82:707, 1952.	rats 880r & 1000r	Decrease in ATP in liver 1st day, rising on 4th day. No change in phosphate distribution in kidney. Fasting p exposure - decreased rate of glycogenolysis. Increased sulphydral content in liver. Little change in cytochrome oxidase content.

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
68.	Smith, E.D. et al. Physiological & biochem. studies p massive irradiation Rad. Res. 11:198-205, 1959.	450-500r/min continuous, rats, mice, hamsters bats, chickens, frogs & lizards	All died after 100,000r! Immediate decrease in rectal temperature in homotherms. No significant alterations in enzymes.
69.	Soeda, H. Effect of irradiation on liver function in pts. with malignant tumors. Nippon. NSA 8/31/61, P. 2667	86 humans local exp.	Tests: Kunkel zinc sulfate, sublimate turbidity, BSP bilirubenia, S. cholinestorase. Exposure over abdomen - unfavorable reaction on liver function.
70.	Spoerl, E. & Quiner, W. Glucose uptake & dissimilation by irradiated, starved & division inhibited yeast. Report # 486 Army Med. Res. Lab. Fort Knox	yeast	A concurrent lower rate of glucose uptake of un-irradiated, starved cells as compared to irradiated cells. Discussion - possibility that membrane is site of cellular change involved.
71.	Staukovic, F. Effect of TBI in insulin resistance of fasting rats. Nature 184: Suppl.32, 1816, 12/59	rats 600r	6 of 20 rats survived insulin shock compared to 20 of 20 controls. Decrease in hexokinase activity was also observed.
72.	Stocken, L.A. Observations on biochemical effect of radiation. Rad. Res. Suppl. 1, 53-57, 1959 (summary of reports by different investigators)		II. Enzymes A. ATP - Doubise & Peterson - ATP increase in mouse spleen proportional to dose between 25 - 400r. Similar findings in thymus and for 5-nucleotidase B. Catalase - in vivo & invitro with occasional reduction in activity C. Nucleases - two DNAases are now recognized, DNA I & II, 500r - increase in DNA II in liver. No change in DNA I. III. Oxidation of Phosphorylation A. Mitochondria - Potter & Bethell - Reduced P/O ratios in spleen mitochondria taken from rats exposed to 800r one hr. previously. Van Bekkum - Succinate as substrate, reduced P/O ratio after 100r in spleen & 50r in thymus. At 700r the earliest significant depression in spleen at 2 hrs. No effect in liver at 2 or 4 hrs; nuclear degeneration at 1 hr. with reduced mitotic index at 15 min.

Effect of Radiation - Biochemistry Studies

Title and Reference

Findings

72. Stocken, L.A. continued
cont'd.

III. Oxidation of Phosphorylation
A. continued

Fritz - Niggle - Oxidation of various members of Kreb's cycle
by rat liver mitochondria. 50r inhibited pyruvate oxidation by
30% & citrate by 10 - 15%. 100r before effect on succinate.
0.1r gave 60% inhibition of oxygen uptake 30 to 60 min.

B. Nuclear Phosphorylation - Creasey & Stocken. Only intra-
nuclear phosphorylating systems can separate radiosensitive and
now sensitive tissues. 100r or more suppressed phosphorylation
of spleen, thumus, bone marrow, lymph node nuclei.

C. Concentration of acid soluble phosphates - levels of ATP and
other nucleotides seem to depend on both the tissue and time after
irradiation. Rat thymus - no change in P32 specific activity,
concentration of inorganic phosphate or ATP @ 1 hr. post 1000r.
Latwok & Mann - Rabbit marrow, marked fall in ATP concentration
Rat spleen - no change at 4 hrs. after 700r. At 24 hrs. or 3 days
there are reduced concentrations in rat liver, rat & mouse spleen
& rat skin. The cytochrome C effect may reflect damage to the
mitochondria with a consequent reduction in phosphorylation.

IV. Nucleic Acid. Comprehensive review: 1. Hevesy & Forsberg.
Proc. 3rd Internat. Congr. Biochem. Brussels 1955, 479, 1956.
2. Hevesy, G. Brit. J. Radiol. 29:465, 1956. 3. Evers, M.
Handbruch der Protoplasmaforschung (L.V.Heilbrunn & F. Weber,
eds. p. 1-241). Springer, W. 1957. 4. Kelly, L.D. Prog. in Bio-
phys. & Chem. 8:144, 1957. Reduced amounts of DNA & RNA have
been observed in certain tissues 3 hrs. post irradiation, but changes
are most likely due to disappearance of cells.
Conclusion: A. Howard - Ciba Foundation Symposium, ionizing
radiation & cell metabolism 196, 1956: "Most experimental results
can be explained as due simply to mitotic delay and cell death
and do not require to invoke a biochemical action of radiation
on DNA synethesis per se."

Effect of Radiation - Biochemistry Studies

	Title and Reference	Organism & dose	Findings
73.	Ueno, I., et al. Eosinophilia developed during x-ray therapy. Vestnik Rentgenol.l Radiol.35:55-58, 1960	humans 250-415r/day 6000-9000r	Local exposures. Tendency to either an absolute or relative increase in eosinophil count.
74.	U. Navolone, Italy. Renal function in irradiated patients NSA 8/31/61, p. NSA p. 2667	humans 2000r partial	The maximum tubular excretion of paminohippuric acid (Tm PAI) is a particularly sensitive & useful measure of renal failure during the early post irradiation period. Changes have been noted p 2000r when part of the field included the renal area.
75.	Vogel, H.H., Frigerio, N.R. & Jordan, D.L. Life shortening in mice p fission neutrons or Co-60 gamma rays at low dose rate	mice	Low dose rate is much less effective in shortening life span than is high dose rate.
76.	Weiner, N. et al. Time trend of hyper-lipoproteinemia p radiation injury. Sch. Aviation Med. Randolph AFB 1955	rabbits local	Elevated plasma lipids 1st week, persisted for several weeks. Lipoprotein curves. Muscle histology & ATP levels show progression of local tissue changes after radiation.
77.	Ibid, Idem. Effect of reduced food intake on hyperlipoproteinemia of local radiation injury.	rabbits	Hyperlipidemia, weight loss. Pair fed controls developed increased plasma lipid & lipoprotein with little weight loss.
78.	Yamamoto, G. Effect of radiation on nuclei acid metabolism. I. Radiosensitivity of various organs. NASA 8/31/61, p. 2666	mice 200-300r	Wt. & nucleic acid content of thymus, spleen, liver & testis were measured. Liver is the most radio-resistant with other organs showing decrease in wt. & nucleic acid content. Nucleic acid metabolism was disturbed in thymus, spleen & testis by small sublethal exposures.

Effect of Radiation - Biochemistry Studies

Summary of Results

Tissue, organ or system	Observations and Changes	Exposure	Organism	Reference
Urine	Decreased urease activity	750,000r	in vitro	24
Kidney	Coproporphyrin (no report)		humans	58
	" no dose relationship	100-1000r	rats	51
	Increased deoxycytidine		rats	60
	Creatine - 20% increase	1500r	dogs	45,46
	17 keto - 17 hydroxy increased		calf	25
	Increased adrenal cortical steroids	750-1000r	pigs, rats	5, 15, 37
	Increased beta-amine isobutyric acid	300-500r	humans	40
	Increased chlorine excretion	750-1000r	pigs, rats	5, 15, 37
	No increase in taurine	300-500r	humans	40
	Decreased aminoethanol	300-500r	humans	40
Intestine	Decreased thiamine absorption	525r	rats	19
Eye	Increased damage		rabbits	38, 41
Synovia	Decreased viscosity & cohesiveness of skin		cockscomb, rat, & human skin	50
Blood cells	Depression - hematopoietic system	300-500r	humans	40
	"	0-20r/day	ducks & mice	17,33
	Increased RBC, Hgb, WBC c shift to rt.	low level	humans	73
	Increased L/S lymph, L+M/s specific type	" "	"	65
	Decreased granulocytes, increased myocytes	" "	"	65
	Decreased DNA synthesis of lymphs		rats	4
	Decreased lymphs	500r	dogs	16
	Increased amines	500r	rats	29
	Decreased RBC fragility	100r/wk	dogs	21
	Increased lymphs with bilobed nuclei	low level	humans	36
Brain	No alteration in enzyme activity	450-500r	several species	68
	No change in permeability	10,000r	rats	62

Effect of Radiation - Biochemistry Studies

Summary of Results

Tissue, organ or system	Observations and Changes	Exposure	Organism	Reference
Whole body	No weight loss	700r	mice	31
	Loss of consciousness	heavy	humans	22
	Early nausea	"	"	22
	No vomiting	up to 100r	"	22
	Diarrhea in 24 hrs. - poor prognosis	heavy	"	22
	Decreased food intake	660r	rats	13
	Survival increase c increasing energy	LD-50	guinea pigs	26
	" c increasing dose rate	700r	mice	31
	" c decreasing energy	634-816r	mice	32
	" c decreasing dose rate	80-250 Kv.	rabbits	75
Spleen	No weight loss	700r	mice	31
	Decreased DNA, H$_2$O, fat, Na, K, Cl	LD-50	rats, rabbits	58
Liver	No weight loss	700r	mice	31
	Protoporphyrin (decreased function)	100r	guinea pigs	47, 48
	Decreased ATP, increased on 4th day	1000r	rats	66,67
	Retarded glycogenolysis	880r	rats	67
	Increased sulfhydral compounds	880r	rats	67
	No change in Na, P, H$_2$O	880r	rats	67
	Slight increase in cytochrome oxidase	880r	rats	67
	Increased P^{32} uptake in RNA	880r	rats	67
	Decreased function (several tests)	700r	rats	43
	Increased glycogen (6 to 10 times)		humans	69
	Decreased DNA, water, fat Na, K, Cl	200r	rats	57
	Oxidation & phosphorylation	LD-50	rats, rabbits	59
		(see summary)		72
Testis	No weight loss	700r	rats, rabbits	31
	Wt. loss, same with high & low dose		mice	9
	Increased incidence of testicular lesions	57-154 rep	monkeys	61

Effect of Radiation - Biochemistry Studies

Summary of Results

Tissue, organ or system	Observations and Changes	Exposure	Organism	Reference
Plasma	No lethal toxin identified	1000-20,000r	rats	53
	Increased clotting time, clot retraction time, bleeding time, fibrin time	low levels	humans	39
	Isolation of heparin-like compound		dogs	3
	Increased sugar (10 fold) at 24 hours	200r	rats, yeast	30,44,57,70
	Impaired glucose utilization	250r, 500r	guinea pigs, rats	34
	Increased transaminase at 24 hrs.	700r	rabbits	1,14,66
	Decreased balance of Na, K, Cl	660r	rats	13
	Shift in lipoproteins correlated to death	835r	rabbits	77,56,76
	Decrease beta globulin	330-990r	chickens	52
	Decreased antibody formation & immunity c little effect on acquired immunity	summary of data		64
	Increased enzymes activity	0.3-300r/wk	rats	35
	Increased NPN, A/G, Cl, cholesterol	200r	rats	44
	Creatine - 3 to 4 fold increase	1500r	rats	5
	Protein - (massive, lethal, & sublethal, changes comparable)		in vitro	54
Lymph of thoracic duct	Decreased lymphs by 18%, T. Protein, keratin	500r	rats & dogs	16, 29
	Increased amines, NPN, uric acid			
Kidney	No changes in phosphate	1000r	rats	67
	No change in sulfhydral, Na, por. H_2O	880r	rats	67
	Decreased P^{32} excretion	50-8000r	guinea pigs	48
	Decreased DNA, H_2O, fat, Na, K, Cl	LD-50	rats, rabbits	58, 27, 28
	Decreased renal excretion of PAI	2000r		
		partial	humans	74
Pancreas	Shift in beta-alpha cell ratio	250r	guinea pigs	2

Effect of Radiation - Biochemistry Studies
Summary of Results

Tissue, organ or system	Observations and Changes	Exposure	Organism	Reference
Thymus	Decreased ADP, GTP, TTP, DPN @ 30 min. + AMP, ATP, CTP, UTP @ 7 hrs.	800r	rats	8, 78
	Decreased uptake P32, TTP, AMP @ 30 min.+ ATP, CTP, GTP, UDP @ 7 hrs.			
	Decreased uptake P32 in RNA	800r	rats	8
	Increased ATP	25-400r	mice	72
	No change in inorganic P @ 1 hr.	1000r	rats	72
Adrenal glands	No change in noradrenalin up to 6 hrs. decreased to 0 by 76 hrs.	800r	rats	55,71
	Decreased adrenalin	1770r	rabbits	55,71
	"	1600r	rats	55,71
	No change in either	30,000r	cats	55,71
Thyroid	Increased thyroid activity	1,000r	rats	11

Figure 4.

EFFECT OF IONIZING RADIATION ON LIVING TISSUE

Summary of Investigations

Disturbance at the Molecular Level
1. Sulfhydryl compounds
2. Splitting of H_2O to form
dioxides, peroxides, etc.

Inorganic Radicals (short lived)

Organic Radicals

Damage to Biological Structure
R.B.C. Fragility
Thiamine Absorption

Preprophase Arrest
1. (H^3) thymidine
2. (P^{32}) incorporation
3. (Fe^{59}) uptake
4. Cholinesterase

Cell Division
Mitotic Index

Metabolic Disturbances

Genetic
unbalance

I. Enzymes
   Compound F
   Transaminase
   RNAase
   ATPase
   DNAase
   Catalase
   S-cholinesterase

II. Amino acids, Amines,
   Nucleic acids
   B-aminobutyric acid
   Creatine, creatinine
   Biogenic amines
   2-amino ethanol
   Deoxycytidine
   RNA & DNA
   Bilirubenia

III. Mineral metabolism
   Sodium
   Potassium
   Chlorides
   Iron
   Iodine

IV. Proteins
   T. Nitrogen
   A/G ratio
   Lipoproteins
   Mucopolysaccharieds
   B-globulins
   Immunochemistry
   Protoporphrin metabolisr

V. Steroids
   17 keto - 17 hydroxy

VI. Others
   Carbohydrate metabolism
   Glycogen
   Glucose
   Cholesterol
   Renal function
   Kunkel zinc sulfate
   Sublimate turbidity
   BSP
   etc.

Mutation   Morphological Change   Partial or complete recovery
   Chromosome   Cataract   Bone marrow depression
      abberation   Vascular collapse   G.I. tract injury

Sterilization   Differentiation
                                    1. Ratio of cell types
                                    2. Rebuck test

Death
   1. Single cell type - hematopoietic system, testis, intestinal mucosa
   2. Organ - systemic effects, necrosis, RISA absorption, etc.
   3. Organism - survival time, increased aging

It is possible that the areas of need (outlined on page 20) may be fulfilled by means of an over-all, coordinated program with all participants using a common, comprehensive protocol of procedure. Such a protocol would include dosimetry studies to achieve uniform distribution of irradiation, hematology, biochemistry, and physiology tests, and observations on subjective response. With the accumulation of sufficient material, all data might be evaluated by computer methods to assure reliability of analysis.

If a well coordinated program failed to provide the needed information, then it seems likely that only relative quantitation is possible. While prediction of effect of radiation in a small volume of tissue is possible, prediction becomes more difficult as volume of tissue increases. Measurement of change and function of complex biologic systems presents many problems even under normal conditions so it is not surprising that effect of total body irradiation, which involves all tissues, organs and systems undergoing constant changes of wear and repair, would present inexplicable problems in measurement. If effect of total body irradiation is non-specific because of the many factors involved, then evaluation of the organism as a whole may provide information overlooked in the search for a specific biologic index. Evaluation of the whole individual would relate amount of total body irradiation with performance, degree of incapacitation and survival. Based on clinical observations that take into account biologic and physiologic responses, it would permit prediction of effect of exposures ranging to the LD-100. Such a prediction was presented as an addendum to the progress report of January 31, 1963. This prediction may not be the final answer but, because it represents the "best" answer that can be given from an appraisal of presently available data, it is included in its entirety in this final report.

A PREDICTION OF THE EFFECT OF TOTAL BODY IRRADIATION
BASED ON CLINICAL OBSERVATIONS

The essential problem posed by the possibility of massive irradiation of the human is to predict the lethal dose, the dose at which immediate but temporary incapacity will occur, and the dose at which delayed but significant injury will occur. The possible answers proliferate as probable qualifying factors are introduced, e.g.,

1. Unless there is a uniform concept of dose and dosimetric procedures, gross discrepancies in estimates of dose-effect are to be expected.
2. The effects of irradiation are not specific and are apt to be confused with other types of injury,--mental, physical or biochemical.
3. The effect of the accidental or catastrophic irradiation exposure will vary with the geographic, geometric and anatomic relations that may be unsuspected or, at least, difficult to reconstruct.
4. Age, general health and competing disease will modify the response of the individual. Prediction of dose-effect is possible for individuals or for uniform groups of individuals but a very wide rante of dose-effects is to be expected in the general population.
5. Prediction of dose-effect in humans must be based upon the controlled circumstances of clinical use of radiotherapy where uniformity of radiation throughout the body may be sought although this is unlikely ever to be achieved in accidental or catastrophic exposure.
6. Morbidity and mortality will clearly depend upon medical assistance whether this be merely supportive or actually specific.

Nevertheless, some simple expression of the hazard of radiation is necessary and a prediction can be offered if an allowance of $\pm$ 3 d B is granted.

The following predictions are based on a hypothetical group of adult males between 30 and 50 years of age, with training or education to permit a basic understanding of radiation and biology, physically and mentally conditioned to work in the field of radiation hazards and free of any previous physical or chemical trauma likely to impair tolerance to radiation. The dose is expressed as the amount of radiation measured in air at the site where the individual is exposed. The geometry of exposure and quality of irradiation is assumed to be such as to produce a uniform distribution through the entire body.

"Acute incapacity" means uncertain ability to carry out crucial mental or physical tasks.

"Partial disability" means ability to carry out mental tasks and sedentary work but vulnerable to stress, e.g., severe physical exertion, infection or further irradiation.

The problems of single brief exposure and of protracted exposures are dealt with separately.

Table 15
Predicted Effect
of
Single Exposures of Total Body Irradiation

Amount of Irradiation	Acute Incapacitation	Partial Disability	Blood Counts (% of normal)	Time to Recovery
50r	0	0		
100r	0	0	75%	60 days
200r	0	0	50%	60 days
300r	1 - 6 hrs. (minor)	0	25%	60 days
500r (no deaths anticipated below this level)	1 - 12 hrs.	2 - 3 mos.	5%	90 days
700r (LD-50 range)	1 - 72 hrs.	6 mos.	1%	120 days
900r (LD-100 range)	2 mos.	12 mos. (if any survivors)	<1%	1 year (if any survivors)

The prediction of the effects of protracted total body irradiation is less readily described in simple numerical values for dose and time.

The rule to be anticipate is that, taking values for single exposures of the order of one hour as a standard, the effect of protraction will be (A) to obscure the time of onset of signs or symptoms due to delayed and insidious development , and (B) to prolong the recovery period due to: 1) increased time required for accumulation of an effective amount of irradiation, 2) inhibition of the repairative response, 3) vulnerability to infection and other forms of stress during the period when repairative response is impaired.

It is evident that at some low level of protracted exposure, no detectable effect will occur and at some high level of protraction falling short of lethality, the above description will be clearly applicable. Intermediate exposures will be difficult to define with precision.

Table 16

Predicted Effect
of
Protracted Total Body Irradiation

Amount of Radiation and Duration of Exposure	Symptoms		Blood Counts	
	Acute Incapacitation	Partial Disability	% of normal	Recovery time
100r in 1 wk.	0	0	90%	
in 1 mo.	0	0	90%	
in 1 yr.	0	0	0	
200r in 1 wk.	0	0	75%	
in 1 mo.	0	0	50%	
in 1 yr.	0	0	0	
300r in 1 wk.	0	Day 15-50	50%	3 mos.
in 1 mo.	0	Day 30-90	25%	4 mos.
in 1 yr.	0	0	0	
500r in 1 wk.	Day 7-10	Day 10-90	20%	4 mos.
(MLD range) in 1 mo.	Day 25-35	Day 35-150	1%	6 mos.
in 1 yr.	0	Mos. 10-24	50%	4 mos.
700r in 1 wk.	Day 5-30	Day 30-150	1%	6 mos.
(LD-50 range) in 1 mo.	Day 15-50	Day 50-200	<1%	12 mos.
in 1 yr.	0	Mos. 10-24	25%	4 mos.
900r in 1 wk.	Day 4-30	Mos. 1-6	1%	1 yr.
(LD-90 range) in 1 mo.	Day 10-50	Mos. 2-12	<1%	2 yrs.
in 1 yr.	0	Mos. 8-24	10%	1 yr.

The above values are predictions, not conclusions.

They are offered as "best answers" to current conjecture based on total experience in clinical radiotherapy.

The values below 300r are firm estimates.

The values below 500r are safe estimates.

The reason for continued clinical investigation is to accumulate further observations and develop more reliable tests to confirm or alter the impressions in high dose ranges.

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